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27. (First time amended) A kit allowing the interchangeable use of a DNA library in more than one application, comprising:

a plurality of first PCR oligonucleotide primers, each of said first PCR primers having a first region [homologous with] which is capable of hybridizing to the first common sequence used in the construction of said DNA library, and a second region homologous with a first region of a vector required for a particular application;

a plurality of second PCR oligonucleotide primers, each of said second PCR primers having a first region which is capable of hybridizing to [homologous with] the second common sequence used in the construction of said DNA library, and a second region homologous with a second region of a vector required for a particular application; and instructions for use.

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REMARKS

The Office Action

Claims 2-27 have been rejected under 35 U.S.C. §112, second paragraph. Claims 1-4 and 6-27 have been rejected under 35 U.S.C. §103(a).

Pending Claims

Claims 28-31 have been canceled as being directed to a non-elected invention without prejudice to prosecution in one or more divisional applications.

Claims 2, 3, 11-14, 22, 26, and 27 have been amended. Upon entry of this amendment claims 1-27 will be pending in the application. No new subject matter has been added.

Claims 2, 3, 11-14, 26, and 27 have been amended to replace the phrase "homologous with" by "capable of hybridizing". Claim 22 has been amended to replace the term "derived" by "obtained".

In view of the foregoing claim amendments and the following remarks, it is respectfully submitted that the application is in condition for allowance.

Submission of Sequence Listing

In response to the "Notice to Comply with the Requirements for Patent Applications Containing Nucleotide and/or Amino Acid Sequence Disclosures", Applicant submits herewith a diskette containing a computer readable form of the Sequence Listing, a paper copy of the Sequence Listing and a Verified Statement that the contents of the paper and computer readable copies are the same, and do not include new matter, in accordance with 37 CFR 1.821(f).

Rejection of Claims 2-27 under 35 U.S.C. §112, Second Paragraph

In paragraphs 4a-b of the outstanding Office Action, the Examiner has rejected claims 2-27 under 35 U.S.C. §112, second paragraph, "as being indefinite for failing to particularly point out and distinctly claim the subject matter which applicant regards as the invention". Each of the basis for the rejection is addressed individually below:

a. Claims 2-27 are rejected as being 'vague and indefinite because it is unclear whether the language "said first primers having a first region homologous with the first common sequence of the nucleic acid molecule" means that the first region of the said first primers is complementary to the first common sequence of the nucleic acid molecule'.

This rejection has been met by amending the rejected to replace the language objected to by the Examiner with the phrase "capable of hybridizing". Thus, for example, claim 2, as amended, is directed to a method of preparing a plurality of nucleic acid inserts using a first and a second primers which are capable of hybridizing to the recited target nucleic acid inserts and then amplifying such nucleic acid inserts by using, e.g., polymerase chain reaction.

b. Claim 22 is rejected as being 'vague and indefinite because of the language "is derived from"'.

This rejection has been met by amending claim 22 to replace the language "derived from" with "obtained from".

In view of the foregoing claim amendments and the following remarks, it is respectfully submitted that the application is in condition for allowance.

Rejection of Claims 1-4 and 6-27 under 35 U.S.C. 103(a)

In paragraphs 5-8 of the outstanding Office Action, the Examiner has rejected claims 1-4 and 6-27 under 35 U.S.C. 103(a) as being unpatentable over Jorgensen (US 5,925,544) in view of Fraser et al. (US 4,870,023) or Liu et al. (US 5,928,868). Each of these rejections is addressed individually below.

I. In one aspect of this rejection, the Examiner has rejected claims 1-4, 6-12, 15-22, 25 and 27 as being obvious over Jorgensen. In particular, the Examiner asserts that:

One of ordinary skill in the art at the time of the instant invention would have been motivated to apply the reference of Jorgensen to construct a DNA library because Jorgensen suggest that a cDNA library may prepared (See column 7, lines 40-62) and the method has obtained an increased number of genomically integrated copies of the DNA sequence desired (See the Abstract). An artisan of ordinary skill in the art at the time of the instant invention would have made the kit as claimed including all ingredients which are used for practicing the method because it was routine practice in the art at the time of the instant invention. It would have been prima facie obvious to carry out the method as claimed.

This portion of the rejection is respectfully traversed.

Claims 1 and 3-26 are drawn, *inter alia*, to methods of constructing a DNA library *in vivo*, by introducing, into a plurality of host cells, (i) a vector having a first and a second region, and (ii) a nucleic acid insert having a first region homologous with said first region of the vector and a second region homologous with said second region of the vector, and allowing homologous recombination and gap repair between the vector and the nucleic acid insert to occur. Claim 2 is directed to methods of preparing the aforesaid nucleic acid insert molecules. Claim 27 is drawn to a kit allowing the interchangeable use of a DNA library in more than one application comprising the recited PCR primers.

The Jorgensen reference discloses a method for amplifying *in vivo* a DNA sequence present in the genome of a cell (designated as B), by integrating a construct comprising the structure C-M-A-D, in which both A and C are homologous with a genomic DNA fragment either flanking or overlapping the DNA sequence B. The sequence C is located in the opposite end of the sequence B compared to A. D represents a DNA sequence which is homologous with a genomic fragment located distal for C as compared to B, and M represents a DNA sequence

encoding a selection marker. The structure A-B-C-M-A-D is created upon integration of the construct into the genome. Selection pressure is used to enrich for cells having an increased number of genomically integrated copies of the DNA sequences B and M.

The Jorgenson reference fails to teach or suggest the claimed methods. As described above, this reference discloses a selection method for enhancing the number of cells containing genomically integrated copies of the M marker gene in the vicinity of a B DNA sequence present in the genome. The C-M-A-D construct in the Jorgenson reference is used to target the integration of the M marker sequence at a site near a DNA sequence of interest (the B DNA sequence) into the genome.

The claimed invention is directed to a completely different method from the one disclosed by the Jorgenson reference. The invention is drawn to a method for generating a DNA library (a collection of constructs having different inserts) by using host cells' homologous recombination and gap repair mechanisms for covalently linking the aforesaid nucleic acid insert (containing a DNA library) to the vector. Therefore, the present invention is directed to a method of generating a plurality of vectors containing the aforesaid inserts in a cell.

There is no teaching or suggestion in the Jorgenson reference regarding the use of the constructs described therein to generate a DNA library *in vivo* in the host cell's genome, let alone in a vector molecule. The passage from this reference referred to by the Examiner as describing the preparation of a cDNA or genomic library from the organism describes simply how to identify DNA sequences A, C and D, which overlap or flank the B sequences to generate the integrating C-M-A-D construct. In particular, the Jorgenson reference provides at column 7, lines 34-62:

From the above disclosure it will be understood that the DNA sequences A and C may be homologous with any genomic sequence overlapping or flanking the DNA sequence B. When the DNA sequence B is a gene, the DNA sequence A or C may advantageously be homologous to a full or partial promoter sequence upstream of the coding part of the DNA sequence B. An example of such construct is shown in Example 1 hereinafter.

The DNA construct used in the method of the invention may be synthesized through a series of genetic manipulations employing methods and enzymes known in the art. Typically, each of genomic sequences with which the DNA sequences A, C and D are to be homologous are identified by conventional DNA analysis methods.

For instance, a cDNA or genomic library may be prepared from the organism in question and the DNA sequence B to be amplified identified therein. When at least a part of the DNA sequence B is known, the DNA sequence B may be identified by screening for positive clones by conventional hybridization procedures, e.g. using oligonucleotide probes synthesized on the part of the DNA sequence B in accordance with standard techniques (cf. Sambrook et al., 1989), or more preferably, by use of polymerase chain reaction (PCR) using degenerate oligonucleotide probes prepared on the basis of the known part of the DNA sequence B. For instance, the PCR may be carried out using the techniques described in U.S. Pat. No. 4,683,202 or by R. K. Saiki et al. (1988).

Therefore, the mention of cDNA or genomic library in the Jorgenson reference relates to ways of identifying the A, C and D DNA sequences to be used in the integrating construct, as opposed to methods for constructing an *in vivo* DNA library, as presently claimed.

The Examiner further states that the Jorgensen reference discloses at column 7, lines 62-67 the screening of a cDNA when "an expression product is known for an activity of the product and thereby identify a clone from which the activity is expressed".

As mentioned above, the mention of cDNA or genomic library screening refers to methods of isolating portions of the DNA B sequence (as well as, flanking A, C and D DNA sequences) to generate the insertion constructs. This refers is completely silent regarding methods for constructing an *in vivo* DNA library, as presently claimed.

In sum, the Jorgenson reference fails to disclose a vector/insert combination used to generate DNA libraries *in vivo*, as recited by the present claims. Moreover, there is no teaching or suggestion in this reference that would have led one of ordinary skill in the art to use the vectors and inserts recited in the claims to generate an *in vivo* DNA library. The teachings in Jorgensen are directed to a completely different method for enriching for cells containing a B sequence of interest. Accordingly, no *prima facie* case of obviousness has been established for the claimed methods.

II. In another aspect of this rejection, the Examiner has rejected claims 3 and 13-14 under 35 U.S.C. 103(a) as being unpatentable over Jorgensen (U.S. 5,925,544) in view of Fraser et al. (U.S. 4,870,023). In particular, the Examiner states that:

One of ordinary skill in the art at the time of the instant invention would have been motivated to combine the references of Jorgensen and Fraser et al. for a reasonable expectation of success because Jorgensen suggests that a cDNA library may prepared (See column 7, lines 40-62) and the method has obtained an increased number of genomically integrated copies of the DNA sequence desired (See the Abstract) and the method of Fraser et al. is useful for express vector (See column 3, lines 2-5). It would have been prima facie obvious to make the adapter as claimed.

Applicant respectfully traverses this rejection.

Claims 3 and 13-14 are drawn to methods of constructing a DNA library *in vivo*, by generating a plurality of nucleic acid inserts having the recited structure; introducing into a plurality of host cells a vector having a first and a second region and the aforesaid nucleic acid insert, and allowing homologous recombination and gap repair between the vector and the nucleic acid insert to occur.

The teachings of the Jorgensen reference were distinguished from the claimed invention in section I above. The present invention is directed to a method of generating a plurality of vectors containing the aforesaid inserts in a cell, thereby producing a DNA library. In contrast, the C-M-A-D construct in the Jorgensen reference is used to target the integration of the M marker sequence at a site near a DNA sequence of interest (the B DNA sequence) into the genome, and thus using selection pressure to enrich for cells containing the integrated construct. The teachings in Jorgensen are directed to a completely different method than the one presently claimed. As described above, the mention of a cDNA library in Jorgensen relates to ways of identifying the A, C and D DNA sequences to be used in the C-M-A-D integrating construct. There is no teaching or suggestion in that reference of methods for constructing an *in vivo* DNA library, as presently claimed.

Fraser et al. fail to make up for the deficiencies in the Jorgensen reference. The Fraser et al. reference discloses a recombinant baculovirus construct which encodes fusion polyhedrin proteins capable of forming occlusion bodies containing foreign peptides. The recombinant baculoviruses are formed by insertion into or replacement of regions of the polyhedrin gene that are not essential for occlusion body formation with exogenous DNA fragments.

There is no teaching or suggestion in Jorgensen or the Fraser et al. reference, alone or in combination, describing the vector/insert combination recited by the present claims and its use to

generate an *in vivo* DNA library. The teachings in Jorgensen are directed to a completely different method for enriching for cells containing a B sequence of interest. The baculovirus construct disclosed by Fraser et al. does not supply the elements missing from the primary reference.

Accordingly, the Examiner has failed to establish a *prima facie* case of obviousness for the claimed methods. The Examiner is respectfully requested to reconsider and withdraw this rejection.

III. In yet another aspect, the Examiner has rejected claims 3, 23-24 and 26 under 35 U.S.C. 103(a) as being unpatentable over Jorgensen (U.S. 5,925,544) in view of Liu et al. (U.S. 5,928,868). In particular, the Examiner states that:

One of ordinary skill in the art would have been motivated to combine the references of Jorgensen and Liu et al. for a reasonable expectation of success because the method of Jorgensen has obtained an increased number of genomically integrated copies of DNA sequence desired (See the Abstract) and Jorgensen suggests cDNA may be prepared from the organism (See column 7, lines 49-51) and the method of Liu et al. is rapid to a small molecule (See column 2, lines 25-30). It would have been prima facie obvious to carry out the method as claimed.

Applicant respectfully traverses this rejection.

Claims 3 and 23-24 and 26 are drawn to methods of constructing a DNA library *in vivo*, by generating a plurality of nucleic acid inserts having the recited structure; introducing into a plurality of host cells a vector having a first and a second region and the aforesaid nucleic acid insert, and allowing homologous recombination and gap repair between the vector and the nucleic acid insert to occur. Claims 23-24 and 26 are drawn to methods which further include introducing into a host cell hybrid proteins such that a two hybrid system is performed.

The teachings of the Jorgensen reference were distinguished from the claimed invention in sections I and II above.

Liu et al. fail to make up for the deficiencies in the Jorgensen reference. Liu et al. disclose methods for characterizing small molecules, or for identifying protein targets to which small molecules bind, by introducing, into a cell, a hybrid ligand comprising a small molecule

and a ligand having specificity for a predetermined target; wherein said cell contains a first and second expression vectors. Each expression vector includes a DNA sequence that expresses a hybrid protein linked to a transcriptional modulator domain, the first expression vector comprises a hybrid protein of the predetermined target (to which the aforesaid ligand binds) fused to a DNA binding domain, and the second expression vector comprising a random protein expressed by a cDNA library fused to a transcriptional activator domain. The host cell further includes DNA binding sequences recognized by the DNA binding domain of the predetermined target protein upstream of a reporter gene. The predetermined target portion of the hybrid protein is brought within close proximity of the reporter gene via its DNA binding domain. This predetermined target portion, in turn, binds to the portion of the ligand having specificity for the predetermined target. Transcription activity is activated upon interaction of the hybrid protein expressing the random protein with the small molecule arm of the hybrid ligand.

The invention of claims 23-24 and 26 is directed to a method of generating a DNA library *in vivo* as described. The circularized vectors produced by the host cell further include a transcription factor activator domain. The host cell further includes a test protein fused to a transcription factor DNA binding domain and a reporter gene under the transcriptional control of the DNA binding site recognized by the transcription factor DNA binding domain. Once the DNA library is generated in the host, the encoded library proteins will initiate transcription activity only when these proteins interact with the test protein.

The combination of Jorgenson and Liu et al. fails to provide the elements required by the claimed methods. First, the combination of these references does not teach or suggest the vector/insert combination used to generate the DNA libraries *in vivo* required by the claims. Second, the hybrid system described by Liu et al. is an assay for small molecule screening, as opposed to a method for identifying interacting proteins expressed by the *in vivo* generated libraries. Therefore, the combination of the references cited by the Examiner does not provide all of the elements recited by the claims.

Even assuming *arguendo* that the combination of these references provides the claimed element (which Applicant completely disagrees), there is no motivation in the references cited to combine the homologous recombination system disclosed by Jorgensen with the small molecule screening/two hybrid assay described by Liu et al. The Examiner has tried to reconstruct the

present invention by piecing together the elements of the claimed invention using the present disclosure as a blueprint. *See e.g., Interconnect Planning Corp. v. Feil* 774 F.2d 1132 (“when prior art references require selective combination to render obvious a subsequent invention, there must be some reason for the combination other than the hindsight obtained from the invention itself”).

In view of the foregoing reconsideration and withdrawal of this rejection is respectfully requested.

**SUMMARY**

The present claims are in condition for allowance.

Amendment and cancellation of these claims should not be construed as an acquiescence to the Examiner's rejection. These amendments and cancellations are being made solely for the purpose of expediting prosecution of the above-identified application. Applicant reserves the right to pursue the claims in this application or another application.

If a telephone conversation with Applicant's Attorney would expedite the prosecution of the above-identified application, the Examiner is urged to call Applicant's Attorney at (617) 542-5070.

Please charge the required fee for an extension of time, and any other charges or credits to Deposit Account No. 06-1050.

Respectfully submitted,

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